



Levels of Minerals in *Abelmoschus esculentus* (L.) Moench (Okra) Grown in Zaria Metropolis, Nigeria

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Authors' contributions

This work was carried out in collaboration between all authors. Author MSS designed the study, author SU wrote the protocol and the first draft of the manuscript. Author SZ managed the literature searches, analyses of the study, performed the spectroscopic analysis and author PAE managed the experimental process, author PAE identified the species of plant. All authors read and approved the final manuscript.

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ABSTRACT

This paper was aimed at providing information on the levels of minerals in *Abelmoschus esculentus* (L.) Moench (Okra) grown in Fadaman Kubanni Farms and associated soils, the results were compared with those of the control site (rafin Yashi). The study was design to assess the levels of metals in the *Abelmososchus esculentus* vegetative parts and fruit samples of Fadaman Kubanni and Rafin Yashi. The study was conducted between June 2011 and May, 2013 in the Department of Chemistry, Ahmadu Bello University, Zaria. The standard methods were used for the determination of phosphorous and metal contents. The results were expressed as mean \pm SD of five replicate determinations. The level of significance differences were determined at $p \leq 0.05$. The mean concentration ranges of 213.19 \pm 2.41(FP) to 4456.36 \pm 3.41(CP), 94.80 \pm 12.07(FF) to 3338.65 \pm 6.33 (FS), 153.00 \pm 32.99 (CS) to 356.00 \pm 0.66 (FS), 30.35 \pm 8.33 (FS) to 227.00 \pm 24.19 (FP), 39.17 \pm 15.92 (FS) to 4419.36 \pm 22.56 (FP) and 19.50 \pm 0.01 (CS) to 2241.66 \pm 92.04 mg/100g (FF) were recorded for Ca, Fe, K, Na, Mg and P, respectively. The concentrations of Ca,

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Fe, Mg and Mg were appreciable in the analysed samples, thus it could be used as an alternative source of these elements in the body system to alleviate malnutrition, especially, among the socially economic lower classes of the population in the 3rd world country, such as Nigeria.

Keywords: Mineral level; *Abelmoschus esculentus* (L.) Moench; vegetative part; industrial effluents; soil.

1. INTRODUCTION

Vegetables are those herbaceous plants whose part(s) is (are) eaten as supporting food or main dishes [1]. The utilization of leafy vegetables is part of African cultural heritage and they play important roles in the customs, traditions and food culture of the African household.

Nigeria is endowed with a variety of traditional vegetables and different types are consumed by various ethnic groups for different reasons [2,3]. Vegetables play, important role in human diet as they are important sources of minerals especially, Ca, Fe and a good source of vitamins. Vegetables are the cheapest and the most available source of minerals and essential amino acids. They are included in the meals for their nutritional values. However, some are reserved for the sick and convalescence because of the medicinal properties.

Okra, known as *Abelmoschus esculentus*, is an important vegetable in tropical and sub-tropical areas. In the tender stage, it is very nutritious and the season of availability for its use is long. Its fruits are relished all over West Africa and it provides an important source of Ca, K and other mineral matters which are often lacking in diets of developing countries [4]. Okra which is also known as "Gumbo" or lady's finger is widely consumed as fresh vegetable in both temperate and tropical countries. Although the seed pods are more often used [5], the mature seed is known to have superior nutritional quality.

Fruits and vegetables are important components of healthy and nutritious diets and if consumed daily in sufficient amounts as it could help prevent major diseases such as cardiovascular disease and certain cancers. According to report by WHO [6], low fruit and vegetable intake is estimated to cause about 31% ischaemic heart disease and 11% of stroke worldwide. Also, up to 2.7million lives could potentially be saved each year if fruit and vegetable consumption was sufficiently increased. The recent joint FAO/WHO expert consultation on diet nutrition and prevention of chronic diseases recommended the

intake of minimum of 400 g of fruits and vegetables per day for the prevention of chronic disease such as heart disease, cancer, diabetes and obesity as well as for the prevention and alleviation of several micronutrient deficiencies, especially in less developed countries [6].

Okra proves beneficial in normalizing blood pressure and cholesterol level in human body. The vegetable is good for asthma. The vitamin C present in it is an anti-oxidant and anti-inflammatory which helps to curtail the development of asthma symptoms. Okra is good for atherosclerosis and it is also believed to protect some forms of cancer. Some researchers have shown that eating okra lowers the risk of cataracts, it is also good for preventing diabetes and protects from skin ailments like pimples [7].

Fadaman Kubanni is located very close to industrial estate in Zaria, Kaduna state, Nigeria. Industries discharge their effluents into river Kubanni and these effluents contain some levels of nutrients. Okra is grown around this river throughout the year by the inhabitants using this water. Thus, the research was aimed at determining the mineral contents of *Abelmoschus esculentus* grown in Fadaman Kubanni where effluents from industrial estate are discharged and the results compared with those of the control sites (Rafin Yashi).

2. METHODOLOGY

2.1 Reagents

All the reagents used were of analytical grades and standard solutions were prepared with distilled water. All the glasswares were properly washed with distilled water, rinsed with liquid soap, soaked in 10% HNO₃ and then rewashed with distilled water [8].

2.2 Sample Collection

Five Farmlands were randomly selected for the sampling of soils and plant in Kubanni Farmlands, Zaria and Rafin Yashi, Giwa. The samples of both soils and plants were collected

by coning and quartering method. The soil samples collected from Kubanni Farmlands (study area) and Rafin Yashi (the control site) were coded as FS and CS, respectively. The vegetative part and Okra fruit samples collected from the study area were labeled as FP and FF while those collected from the control site were labeled as CP and CF, respectively.

2.3 Sample Pre-treatment

Soil samples of Fadaman Kubanni and those of the control site (FS and CS) were oven dried, sieved and then kept in clean polythene bags prior to analysis. The collected samples were washed with the distilled water, cut into pieces, air dried and then oven dried. The dried samples were pounded using mortar and pestle and then sieved. The sieved samples were packed in an air tight labeled containers prior to analysis.

2.4 Determination of Mineral Contents of the Samples

2.4.1 Determination of Fe, Ca, Mg, K and Na in soil

1.0g of the soil sample was placed in a 250 cm³ round bottom flask quit – fit pyrex flask and 50 cm³ mixture of hot concentrated HNO₃ and HClO₄ (4:1) was added. The flask was then placed on the hot plate until a clear solution was obtained. The solution was cooled and filtered through acid washed Whatman No.1 filter paper and then made up to a volume of 100cm³ with distilled water [9].

2.4.2 Determination of Fe, Ca, Mg, K and Na in vegetative part and fruit of okra

1.0 g of the pounded plant samples were digested with 20 cm³ of a mixture of HNO₃ and HCl (3:1). The contents were heated on a hot plate at 100 °C for 45 minutes. This was then transferred was still warm through a Whatman No.1 filter paper, the digest were top up to a volume of 100 cm³ with distilled water [10].

2.4.3 Phosphorous determination by vanado-molybdate method

0.50 g each of the soil, vegetative part and fruit samples were weighed into 100 ml beaker and 30 cm³ mixtures of HNO₃, HClO₄ and HCl (65:8:2) was added. The beaker was then placed on the hot plate in the fume cupboard and heated at 150 °C for 1hour. The temperature was

then increased to 235°C until the HNO₃ fumes were driven off. The digest was concentrated to about 10 cm³. The beaker and content were cooled and transferred into 50 cm³ volumetric flask and made up to mark with distilled water. 10ml of the digest was pipette into 50 cm³ volumetric flask and 20 cm³ of distilled water was added and diluted to mark with the distilled water. The mixture was shaken and allowed to stand for 15 minutes for colour development after which the optical density of the solution was read at 420 nm with spectrophotometer 70 [11], the absorbance was recorded and the concentration was extrapolated from the calibration curve.

2.5 Statistical Treatment of Data

The data was statistically analysed using statistical package for social scientist (SPSS) version 12.0 and Microsoft Excel Spread sheet.

3. RESULTS AND DISCUSSION

3.1 Nutrient Contents in Vegetative Part and Fruit of Okra

The concentrations of minerals in the analysed samples are presented in Figs. 3.1 to 3.6, respectively.

3.1.1 Concentrations of calcium

As presented in Fig. 3.1, the concentrations of 617.17, 1281, 213, 4456, 922.47 and 720.89 mg/100g were recorded in the samples of FS, CS, FP, CP, FF and CF, respectively. On comparing the concentration Ca among the samples, the highest concentration was recorded at the vegetative part of the okra sample from the control site while the lowest concentration of this metal was recorded at the FP sample of the sampling point, this clearly indicates that the levels of Ca in the vegetative part of the samples are attributed to other factors such as the nature of the soil and not the industrial effluents. However, the concentration of Ca in the fruit samples at the control site (CF) were lower than those at the sampling site, this was attributed to effect of the industrial waste and bioaccumulation effects. It was reported a concentration range of calcium as 3001-07 to 3205.49 mg/kg in two varieties African pear [12]. Also, a concentration range of 681 to 11650 mg/kg was reported in pulp of grape fruit [13]. These concentration ranges were above the range obtained in this study. Calcium is known for human nutrition in the development and growth of skeletal bone as

well as co-enzymes in metabolic regulations of bio-molecules, it is also a major factor sustaining strong bones and plays a part in muscle contraction and relaxation, blood clotting, synaptic transmission and absorption of vitamin 12. Okra is a good source of calcium which helps to keep strong and lessen the chance of fractures [14]. Calcium containing substances are required by children pregnant and lactating woman for bones and teeth development, the recommended dietary allowance for Ca is 800 mg/day [15]. The concentrations of Ca were significantly different among the samples at $p \leq 0.05$ and were significantly positively correlated among the samples and metals (Appendices I and II), this apparently indicates the common source of the metal ions.

3.1.2 Concentrations of iron (Fe)

As presented in Fig. 3.2, the mean concentrations of Fe in the FS, CS, FP, CP, FF and CF samples were: 3338.65, 1246.65, 562.87, 295.08, 94.80 and 735.81mg/100g, respectively. The highest and lowest concentrations of Fe were recorded in the FS and FF samples as presented in the Figure. This was attributed to the nature of the soil and the bioaccumulative effects. In addition, the concentration of Fe in soil of the control and sampling point were significantly different which signifies the impact of the industrial waste to soil, consequently, the quality of plants that are grown in the this soil is adversely affected. Fe plays an important role in the formation of haemoglobin in the body system and its deficiency leads to anaemia, a disease which is prevalent in developing countries due to poor nutrition

[16,17,18]. Fe is said to be an important element in diet of pregnant women, nursing mothers, infants, convulsing patients and elderly to prevent anaemia and other related diseases [19]. The recommended dietary allowance of Fe is 2-5 mg/day [20]. The concentrations of Fe were significantly different among the samples at $p \leq 0.05$ and were significantly positively correlated among the samples and metals (Appendices I and II). This apparently indicates the common source of pollution of the samples.

3.1.3 Concentrations of magnesium (Mg)

The mean concentrations of Mg in the in the FS, CS, FP, CP, FF and CF samples as presented in Fig. 3.3 were: 39.17, 355.14, 4419.36, 316.50, 551.23 and 63.60 mg/100g, respectively. Generally, the highest and lowest concentrations of Mg were recorded at the FP and FS samples, respectively. This trend was attributed to bioaccumulative effects due to industrial effluents discharge. The recommended daily intake of Mg ranges from 320 to 420 g/day and it plays an important role in protein biosynthesis [21]. Mg plays an important role the stability of the nervous system, muscles contraction and as an activator of alkaline phosphatase. Mg deficiency in the body leads to reduction in the osteoblasts formation in bones [22,23,24]. It can also be used as alternative to Ca in the body [25,26]. The concentrations of Mg were significantly different among the samples at $p \leq 0.05$ and were significantly positively correlated between the samples and metals (Appendices I and II), this apparently indicates the common source of the metal ions.

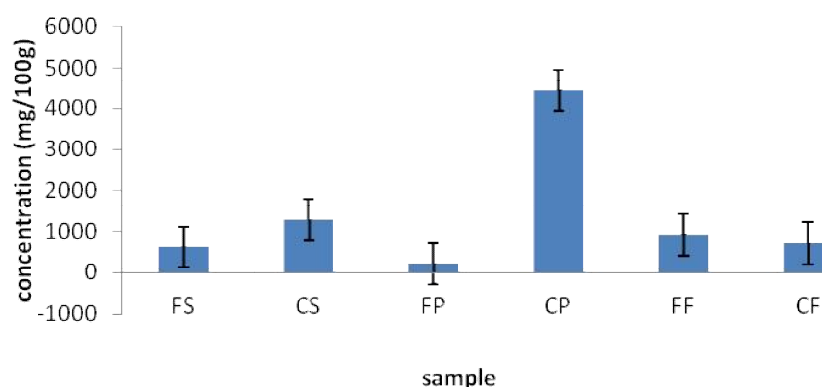


Fig. 3.1. Levels of calcium in the analysed soil, vegetative parts and fruit samples of Fadaman Kubanni and Rafin Yashi

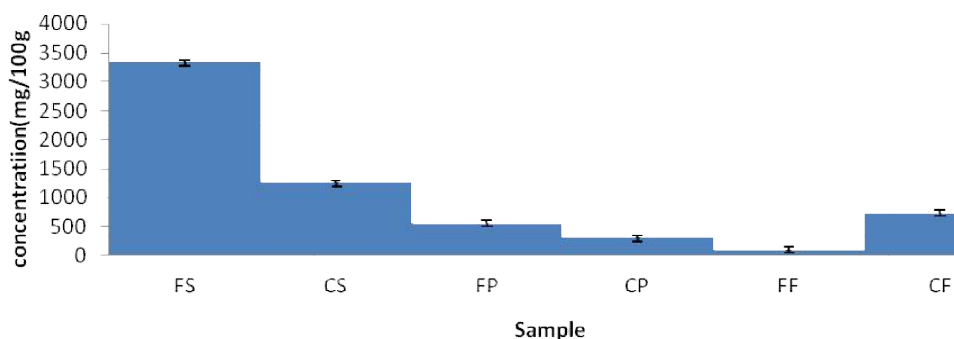


Fig. 3.2. Levels of iron in the analysed soil, vegetative parts and fruit samples of Fadaman Kubanni an

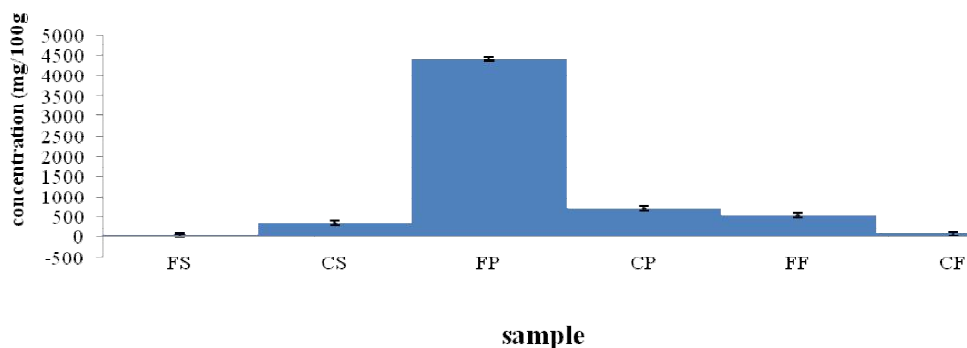


Fig. 3.3. Levels of magnesium in the analysed soil, vegetative parts and fruit samples of Fadaman Kubanni and Rafin Yashi

3.1.4 Concentrations of phosphorous (P)

The levels of phosphorous in the analysed samples of FS, CS, FP, CP, FF and CF as presented in Fig. 3.4 were: 308.85, 19.50, 1800, 933.33, 2241.66 and 2166.66 mg/100g, respectively. Overall, the highest and lowest concentrations of Mg were recorded at the CF and CS samples, respectively. The trend observed in the bioavailability of P was attributed to the nature of the soil in both the control and sampling point and not the industrial effluents discharged. Phosphorous plays important role in the liberation and utilization of energy in animal and vegetable tissues, in the energy metabolism in the formation of sugar and adenosine di- and triphosphate, it is therefore widely distributed in all foods. The normal range of phosphorous in unpolluted soil extract is 0.3 to 8 mg/100g while the normal range in plant materials is 0.05 to 0.3% [27]. The concentrations recorded in this study were above the normal range of phosphorous, this was attributed to other factors such as fertilizer application in addition to

industrial effluents discharged into the river and subsequently used for irrigating the plants. The concentrations of P were significantly different among the samples at $p \leq 0.05$ and were significantly positively correlated among the samples and metals (Appendices I and II), this apparently indicates the common source of the metal ions.

3.1.5 Concentrations of potassium (K)

The levels of potassium in the samples of FS, CS, FP, CP, FF and CF as presented in Fig. 3.5 were: 356, 153, 173.33, 263.22, 351 and 291.96 mg/100mg, respectively. Also, the highest and lowest concentrations of potassium were recorded at the samples of the FS and CS, this was attributed to industrial effluent discharge and bioaccumulative effects. Potassium play an important role in vital cellular metabolism, it catalysis the conversion of adenosine diphosphate (ADP) to adenosine triphosphate (ATP). K is transported to the liver and other body tissues; it also plays a vital role in protein

and carbohydrate metabolism. K- deficiency retards fluid transportation in the body system, it can also leads to increase levels of serum glycerides, obesity which further develops arthrosclerosis and lower blood supply to vital organs and increase the chances of stroke. It was reported that the concentration of 120 mg/day of K can stabilize blood pressure and reduce stroke [28]. The K uptake in plants is dependent upon the soil transport level. High level of clay in soil might lead to low K-uptake by plants since K- would be tightly held in the crystal lattice of the clay and would not easily be released to the environment. The recommended daily intake of K is 4700 mg/day [29]. The concentrations of K were significantly different among the samples at $p \leq 0.05$ and were significantly positively correlated among the samples and metals (Appendices I and II), this apparently indicates the common source of the metal ions.

3.1.6 Concentrations of sodium (Na)

As presented in Fig. 3.6, the concentration of Na in the analysed samples of FS, CS, FP, CP, FF and CF were: 30.33, 36.33, 227.00, 154.66, 55.66 and 75.66 mg/100g, respectively. The highest and lowest concentrations were found in the FP and FS samples, respectively, indicating the impact of the industrial effluent on the quality of soil and plant grown in the study area. However, the concentration of Na in the soil from the control site was higher than the level recorded at the sampling point; this was attributed to other factors such as fertilizer application, application of organic manure etc. Vegetables and fruit diet consist of large amount of Na-salt as required by human body. Na is an essential macro element which has physiological effect in human and animal cellular and metabolic mechanism. The increase level of Na in the body system has direct link to the high blood pressure [30].

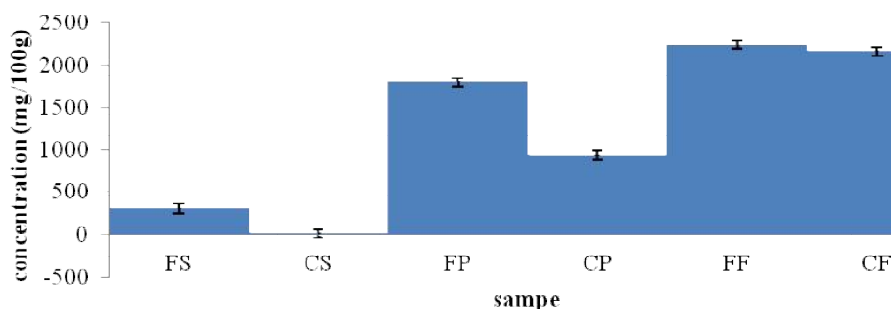


Fig. 3.4. Levels of phosphorous in the analysed soil, vegetative parts and fruit samples of Fadaman Kubanni and Rafin yashi

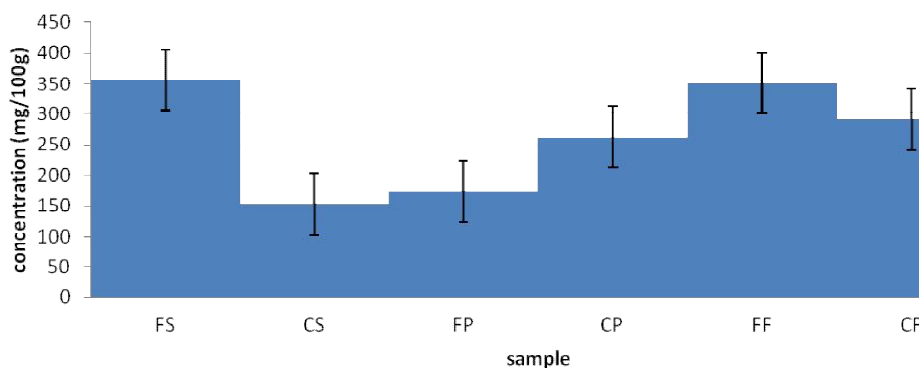


Fig. 3.5. Levels of K in the analysed soil, vegetative parts and fruit samples of Fadaman Kubanni and Rafin Yashi

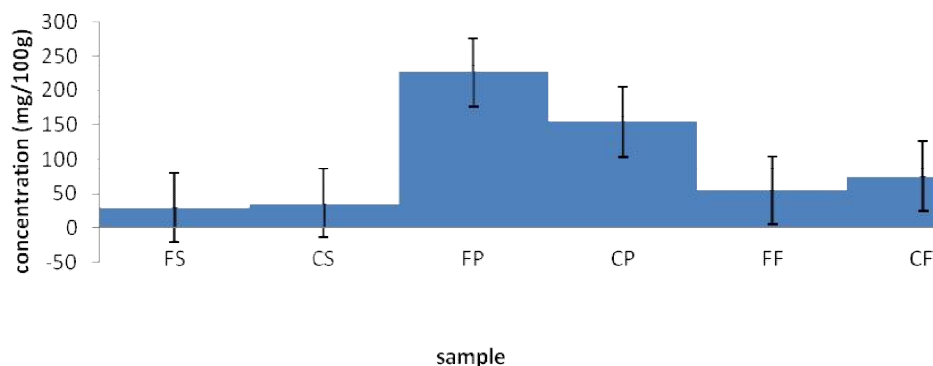


Fig. 3.6. Levels of sodium in the analysed soil, vegetative parts and fruit samples of Fadaman Kubanni and Rafin Yashi

After renal filtration, excessive amount is excreted from the body; the required amount of Na is reserved and re-utilized through blood circulation. Na and K concentration is responsible in total body fluid needed in acid base equilibrium as well as for the osmotic regulation of the body. The recommended daily intake of Na ranges from 2400 to 5175 mg/day [31]. Overall, the bioavailability trend of the mineral elements across the sites was: $P > Ca > Fe > Mg > K > Na$. This clearly indicates that Phosphorous the highest and concentration of Na was lowest among the mineral elements. The concentrations of Na was significantly different among the samples at $p \leq 0.05$ and were significantly positively correlated among the samples and metals (Appendices I and II), this apparently indicates the common source of the metal ions.

4. CONCLUSION

The concentrations of Ca, Fe, Mg and Mg were appreciable in the analysed samples of *Abelmoschus esculentus*, thus it could be used as an alternative source of these elements in the body system to alleviate malnutrition, especially, among the socially economic lower classes of the 3rd world countries, such as Nigeria.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Appendix I. Correlation matrix of mineral contents in the analysed samples across the sites

Sample/Metal	FeFS	FeCS	FeFP	FeCP	FeFF	FeCF	MgFS	MgCS	MgFP	MgCP	MgFF	MgCF	PFS	PCS	PF
FeFS	1.000														
FeCS	1.000**	1.000													
FeFP	1.000**	1.000**	1.000												
FeCP	1.000**	1.000**	1.000**	1.000											
FeFF	1.000**	1.000**	1.000**	1.000**	1.000										
FeCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000									
MgFS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000								
MgCS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000							
MgFP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000						
MgCP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000					
MgFF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000				
MgCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000			
PFS	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000		
PCS	0.985	0.985	0.985	0.985	0.985	0.985	0.985	0.985	0.985	0.985	0.985	0.985	1.000*	1.000	
PF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000
PCP	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
PCF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
PFF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
KFS	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
KCS	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
KFP	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
KCP	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
KFF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
KCF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
NaFS	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.983	1.000**	1.000**	1.000**
NaCS	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.983	1.000**	1.000**	1.000**
NaCP	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
NaFP	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
NaFF	0.981	0.981	0.981	0.981	0.981	0.981	0.981	0.981	0.981	0.981	0.981	0.982	1.000**	1.000*	1.000**
NaCF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
CaFS	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**

CaCS	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
CaFP	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
CaFF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
CaCP	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**
CaCF	0.982	0.982	0.982	0.982	0.982	0.982	0.981	0.982	0.982	0.982	0.982	0.982	1.000**	1.000*	1.000**

** . Correlation is significant at the 0.01 level (2-tailed). * . Correlation is significant at the 0.05 level (2-tailed).

Appendix I. Correlation matrix of mineral contents in the analysed samples across the sites (continued)

Sample/Metal	PCP	PCF	PFF	KFS	KCS	KFP	KCP	KFF	KCF
PCF	1.000**	1.000							
PFF	1.000**	1.000**	1.000						
KFS	1.000**	1.000**	1.000**	1.000					
KCS	1.000**	1.000**	1.000**	1.000**	1.000				
KFP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000			
KCP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000		
KFF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000	
KCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000
NaFS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
NaCS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
NaCP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
NaFP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
NaFF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
NaCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
CaFS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
CaCS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
CaFP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
CaFF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
CaCP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**
CaCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**

Appendix I. Correlation matrix of mineral contents in the analysed samples across the sites (continued)

Sample/Metal	NaFS	NaCS	NaCP	NaFP	NaFF	NaCF	CaFS	CaCS	CaFP	CaFF	CaCP	CaCF
NaCS	1.000**	1.000										
NaCP	1.000**	1.000**	1.000									
NaFP	1.000**	1.000**	1.000**	1.000								
NaFF	1.000**	1.000**	1.000**	1.000**	1.000							
NaCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000						
CaFS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000					
CaCS	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000				
CaFP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000			
CaFF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000		
CaCP	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000	
CaCF	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000**	1.000

Appendix II. Analysis of variance (ANOVA) of the samples across the sites

Metal	Sum of Squares	df	Mean Square	F	Sig.
Ca	35226333.8271	5.0000	7045266.7654	7887.6616	0.0000
Fe	20723393.9030	5.0000	4144678.7806	7918.0810	0.0000
Mg	41436791.6007	5.0000	8287358.3201	10555.1505	0.0000
P	13512695.2876	5.0000	2702539.0575	5062.1922	0.0000
K	109108.5579	5.0000	21821.7116	1242.9998	0.0000
Na	89120.3452	5.0000	17824.0690	5381.1850	0.0000

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